AMENDMENTS TO THE CLAIMS

This listing of the claims replaces all prior listings and versions:

1. (currently amended): A transposon cassette ~~for use in a gram-positive target organism~~, comprising:

a polynucleotide sequence ~~derived from a transposon~~ comprising ~~first and second transposon inverted repeat sequences flanking~~ an internal polynucleotide sequence comprising~~,~~ ~~wherein (i) said internal polynucleotide sequence comprises~~ a first sequence of interest encoding polypeptide sequences present in a first orientation, said first sequence of interest capable of being expressed in a gram-positive target organism and lacking control sequences that are capable of promoting transcription in the target organism; and ~~(ii) said transposon is~~ first and second transposon inverted repeat sequences derived from a gram-positive bacterium, said first and second transposon inverted repeat sequence flanking said internal polynucleotide sequence.

2. (original): The cassette of claim 1, wherein the polynucleotide sequence is derived from TN4001 or TN917.

3. (original): The transposon cassette of claim 1, wherein said Gram-positive bacteria is selected from the group consisting of *Staphylococus spp, Streptococcus spp., Enterococcus spp., Bacillus spp., Clostridium spp., Mycobacterium spp., Corynebacterium spp., Listeria spp., Propriobacterium spp., Micrococcus spp, Lactobacillus pp.,* and *Lactococcus spp.*

4. (original): The transposon cassette of claim 1, wherein the first sequence of interest comprises at least one polynucleotide sequence encoding light generating polypeptide sequences.

5. (original): The transposon cassette of claim 4, wherein said polynucleotide sequence encoding light generating polypeptide sequences is selected from the group consisting of: (a) a polynucleotide encoding *luxA,* and *luxB* gene products; (b) a polynucleotide encoding *luxA, luxB, luxC, luxD* and *luxE* gene products; (c) a polynucleotide encoding *luxY* gene product; and (d) a polynucleotide encoding *luc* gene product.

6. (original): The transposon cassette of claim 4, wherein said polynucleotide sequence encoding light generating polypeptide sequences further comprises at least one Gram-positive ribosome binding site sequence.

7. (original): The transposon cassette of claim 6, wherein said polynucleotide sequence encoding light generating polypeptide sequences encodes *luxA* and *luxB* gene products.

8. (original): The transposon cassette of claim 7, wherein said polynucleotide sequence encoding light generating polypeptide sequences further comprises at least one Gram-positive ribosome binding site sequence upstream of each of the polynucleotide sequences encoding each of the *luxA* and *luxB* gene products.

9. (original): The transposon cassette of claim 7, wherein said polynucleotide sequence encoding light generating polypeptide sequences further comprises a polynucleotide encoding *luxC, luxD,* and *luxE* gene products.

10. (original): The transposon cassette of claim 9, wherein said polynucleotide sequence encoding light generating polypeptide sequences further comprises at least one Gram-positive ribosome binding site sequence upstream of each of the polynucleotide sequences encoding each of the *luxA*, *luxB, luxC, luxD* and *luxE* gene products.

11. (original): The transposon cassette of claim 7, wherein said polynucleotide sequence encoding light generating polypeptide sequences further comprises a polynucleotide sequence encoding the *luxY* gene product.

12. (original): The transposon cassette of claim 11, wherein said polynucleotide sequence encoding light generating polypeptide sequences further comprises at least one Gram-positive ribosome binding site sequence upstream of the *luxY* gene product.

13. (original): The transposon cassette of claim 1, wherein said first sequence of interest further comprises a coding sequence for a selectable marker.

14. (original): The transposon cassette of claim 4, wherein said first sequence of interest further comprises a coding sequence for a selectable marker.

15. (original): The transposon cassette of claim 14, wherein the coding sequence for the selectable marker encodes a polypeptide conferring antibiotic resistance.

16. (original): The transposon cassette of claim 15, wherein said coding sequence for the selectable marker further comprises at least one Gram-positive ribosome binding site sequence upstream said coding sequence for the selectable marker.

17. (original): The transposon cassette of claim 16, wherein said antibiotic is kanamycin.

18. (original): The transposon cassette of claim 4, wherein said first sequence of interest comprises:

(a) a polynucleotide sequence encoding light generating polypeptide sequences comprising a polynucleotide encoding *luxA, luxB, luxC, luxD* and *luxE* gene products, and further comprising at least one Gram-positive ribosome binding site sequence upstream of each of the polynucleotide sequences encoding each of the *luxA, luxB, luxC, luxD* and *luxE* gene products; and

(b) a coding sequence for a selectable marker encoding a polypeptide conferring kanamycin resistance.

19. (original): The transposon cassette of claim 1, wherein said internal polynucleotide sequence further comprises a transposase coding sequence operably linked to a promoter functional in the target organism, said transposase capable of inducing transposition mediated by said transposon inverted repeats, wherein said transposase coding sequence is in a second orientation relative to polypeptide coding sequences of the first sequence of interest encoding polypeptide sequences.

20. (original): The transposon cassette of claim 1, wherein said internal polynucleotide sequence further comprises a transposase coding sequence operably linked to a promoter functional in the target organism, said transposase capable of inducing transposition mediated by said transposon inverted repeats, wherein at least one transcription termination control sequence is interposed between said first sequence of interest encoding polypeptide sequences and said transposase coding sequence which is operably linked to a promoter functional in the target organism.

21. (original): The transposon cassette of claim 19, wherein said first and second transposon inverted repeat sequences, and said transposase coding sequence are derived from *Tn*4001.

22. (original): A vector comprising, (a) a vector backbone and (b) a transposon cassette of claim 19.

23. (original): A vector comprising, (a) a transposon cassette of claim 1, and (b) a vector backbone, said vector backbone comprising a transposase coding sequence operably linked to a promoter functional in the target organism, said transposase capable of inducing transposition mediated by said transposon inverted repeats and wherein said promoter does not affect transcription of any coding sequences in the transposon cassette.

24. (original): The vector of claim 22, said vector backbone comprising an origin of replication that is functional in a target host cell.

25. (original): The vector of claim 24, said vector backbone comprises a Gram-positive origin of replication.

26. (original): The vector of claim 25, wherein said Gram-positive origin of replication is conditional.

27. (original): The vector of claim 26, wherein said conditional Gram-positive origin of replication is temperature-sensitive.

28. (original): The vector of claim 24, wherein said vector backbone comprises a Gram-negative origin of replication.

29. (original): The vector of claim 28, wherein said conditional Gram-negative origin of replication is conditional.

30. (original): The vector of claim 22, said vector backbone comprising an origin of replication that is functional in more than one target host cell.

31. (original): The vector of claim 30, wherein said origin of replication is functional in both Gram-negative and Gram-positive cells.

32. (original): The vector of claim 22, wherein said vector backbone further comprises a selectable marker sequence of interest operably linked to a promoter functional in a target

organism, wherein said promoter does not affect transcription of any coding sequences in the transposon cassette.

33. (original): The vector of claim 32, wherein said selectable marker coding sequence is a polynucleotide sequence encoding a polypeptide conferring antibiotic resistance.

34. (original): The vector of claim 22, wherein said vector backbone further comprises at least one polynucleotide sequence encoding light generating polypeptide sequences operably linked to a promoter functional in a target organism of interest, wherein said promoter does not affect transcription of any coding sequences in the transposon cassette.

35. (original): The vector of claim 34, wherein said transposon cassette contains a polynucleotide sequence encoding light generating polypeptide sequences wherein light generating polypeptide produced from coding sequences within the transposon cassette produce bioluminescence of a characteristic first wavelength that is detectably different from a characteristic second wavelength of bioluminescence produced by the product of the polynucleotide sequence encoding light generating polypeptide sequences contained within the backbone vector.

36. (original): The vector of claim 34, wherein said polynucleotide sequence encoding light generating polypeptide sequences comprises a polynucleotide selected from the group consisting of: (a) a polynucleotide encoding *luxA,* and *luxB* gene products; (b) a polynucleotide encoding *luxA, luxB, luxC, luxD* and *luxE* gene products; (c) a polynucleotide encoding *luxY* gene product; and (d) a polynucleotide encoding *luc* gene product.

37. (original): The vector of claim 22, wherein the vector backbone comprises: (i) a Gram-negative origin of replication; (ii) a Gram-positive origin of replication; and (iii) a selectable marker coding sequence operably linked to a promoter functional in the target organism, wherein said promoter does not affect transcription of any coding sequences in the transposon cassette.

38. (original): The vector of claim 22, wherein said vector backbone is pAUL-A.

39. (original): The vector of claim 22, wherein said vector backbone comprises pE194.

40. (original): The vector of claim 22, wherein said vector backbone comprises pSK.

41. (original): The vector of claim 22, further comprising at least one transcription termination sequence in the vector backbone and adjacent the transposon cassette, such that said transcription termination sequence essentially prevents transcription originating from any promoter present in the vector from reading through into the transposon cassette sequences.

42. (original): The vector of claim 41, comprising two transcription termination sequences in the vector backbone wherein said transcription termination sequences flank the transposon cassette, such that said transcription termination sequences essentially prevent read-through transcription originating from any promoter present in the vector into the transposon cassette sequences.

43. (withdrawn): A method for modifying a microorganism having a genome, comprising transforming said microorganism with the vector of claim 22.

44. (withdrawn): The method of claim 43, wherein said method further comprises culturing the transformed microorganism under conditions that facilitate transposition of the transposon cassette from the vector into the genome of said microorganism.

45. (original): A cell carrying the vector of claim 22.

46. (original): A cell produced by the method of claim 44.

47. (original): A modified host-cell carrying at least one transposon cassette of claim 1, wherein expression of said first sequence of is mediated by a transcriptional promoter endogenous to the target organism.

48. (original): The modified host cell of claim 47, wherein said cell is a Gram-positive bacteria.

49. (original): The modified host cell of claim 48, wherein said Gram-positive bacteria is selected from the group consisting of *Staphylococus spp, Streptococcus spp., Enterococcus spp., Bacillus spp., Clostridium spp., Mycobacterium spp., Corynebacterium spp., Listeria spp., Propriobacterium spp., Micrococcus spp, Lactobacillus pp.,* and *Lactococcus spp.*

50. (original): The modified host-cell of claim 47, wherein the cell exhibits constitutive bioluminescence.

51. (original): The modified host-cell of claim 47, wherein the cell exhibits inducible or repressible bioluminescence.

52. (original): The modified host cell of claim 47, wherein the cell exhibits bioluminescence upon infecting an animal susceptible to infection by the cell.

53. (withdrawn): A method of identifying active host-cell gene promoters, comprising the steps of:

(a) transforming host-cells with the vector of 22;

(b) culturing the transformed host-cells under conditions permitting transposition of the transposon cassette;

(c) screening transformed host-cells for expression of said first sequence of interest encoding polypeptide sequences; and

(d) identifying the active host-cell gene promoter mediating expression of said first sequence of interest encoding polypeptide sequences.

54. (withdrawn): A method of identifying active host-cell gene promoters, comprising the steps of:

(a) infecting a first animal with a microorganism carrying a vector comprising (a) a vector backbone compatible with said microorganism, and (b) a transposon cassette comprising a polynucleotide sequence comprising first and second transposon inverted repeat sequences flanking an internal polynucleotide sequence, wherein said internal polynucleotide sequence comprises a first sequence of interest encoding (i) light generating polypeptide sequences present in a first orientation, and (ii) a polypeptide sequence conferring antibiotic resistance, said first sequence of interest lacking control sequences capable of promoting transcription in the target organism, said internal polynucleotide sequence further comprising a transposase coding sequence operably linked to a promoter functional in the target organism, said transposase capable of inducing transposition mediated by said transposon inverted repeats, wherein said transposase coding sequence is in a second orientation relative to polypeptide coding sequences of the first sequence of interest

(b) selecting, in said animal, for antibiotic resistant transposants;

(c) isolating antibiotic resistant transposants from said animal;

(d) screening *in vitro* said transposants for transposants that do not exhibit constitutive bioluminescence;

(e) infecting a second animal with the transposants that do not exhibit constitutive bioluminescence;

(f) screening for transposants exhibiting bioluminescence *in vivo* upon infection of said second animal;

(g) isolating transposants exhibiting bioluminescence *in vivo* upon infection of the second animal;

(h) identifying the active gene promoter associated with said first sequence of interest in the transposants exhibiting bioluminescence *in vivo* upon infection of the second animal.

55. (withdrawn): A method of screening a compound of interest for pharmacological effectiveness against a microorganism of interest in an animal, comprising the steps of:

(a) infecting a first animal with a microorganism carrying a vector comprising (a) a vector backbone compatible with said microorganism, and (b) a transposon cassette comprising a polynucleotide sequence comprising first and second transposon inverted repeat sequences flanking an internal polynucleotide sequence, wherein said internal polynucleotide sequence comprises a first sequence of interest encoding (i) light generating polypeptide sequences present in a first orientation, and (ii) a polypeptide sequence conferring antibiotic resistance, said first sequence of interest lacking control sequences capable of promoting transcription in the target organism, said internal polynucleotide sequence further comprising a transposase coding sequence operably linked to a promoter functional in the target organism, said transposase capable of inducing transposition mediated by said transposon inverted repeats, wherein said transposase coding sequence is in a second orientation relative to polypeptide coding sequences of the first sequence of interest

(b) selecting, in said animal, for antibiotic resistant transposants;

(c) isolating antibiotic resistant transposants from said animal;

(d) screening *in vitro* said transposants for transposants that do not exhibit constitutive bioluminescence;

(e) infecting a second animal with the transposants that do not exhibit constitutive bioluminescence;

(f) screening for transposants exhibiting bioluminescence *in vivo* upon infection of said second animal;

(g) isolating transposants exhibiting bioluminescence *in vivo* upon infection of the second animal;

(h) infecting a third animal and a fourth animal with transposants exhibiting bioluminescence upon infection;

(i) treating the infected third animal with a compound of interest;

(j) monitoring the treated, infected third animal and the untreated, infected fourth animal for bioluminescence *in vivo*; and

(k) comparing the third and fourth animals to determine whether the compound of interest detectably affects *in vivo* bioluminescence in the third animal relative to the fourth, wherein reducing or eliminating *in vivo* bioluminescence in the third animal relative to the fourth indicates pharmacological effectiveness against the microorganism of interest in the animal.

56. (withdrawn): A method of monitoring the proliferation of a microorganism of interest in a medium of interest, comprising:

(a) transforming the microorganism of interest with a vector comprising (i) a vector backbone compatible with said microorganism, and (ii) a transposon cassette comprising a polynucleotide sequence comprising first and second transposon inverted repeat sequences flanking an internal polynucleotide sequence, wherein said internal polynucleotide sequence comprises a first sequence of interest encoding light generating polypeptide sequences present in a first orientation, said first sequence of interest lacking control sequences capable of promoting transcription in the target organism, said internal polynucleotide sequence further comprising a transposase coding sequence operably linked to a promoter functional in the target organism, said transposase capable of inducing transposition mediated by said transposon inverted repeats, wherein said transposase coding sequence is in a second orientation relative to polypeptide coding sequences of the first sequence of interest encoding polypeptide sequences;

(b) culturing the transformed microorganism under conditions permitting transposition;

(c) screening for transposants exhibiting bioluminescence;

(d) inoculating a sample of the medium of interest with bioluminescent transposants; and

(e) monitoring the medium sample for degree of bioluminescence over time, wherein an increase in the degree of bioluminescence over time is correlated to proliferation of the microorganism in the sample.

57. (withdrawn): The method of claim 56, further comprising

(f) adding a compound of interest to the medium, and

(g) evaluating the effect of the compound on proliferation of the microorganisms.

58. (original): The transposon cassette of claim 15, wherein the coding sequence for the selectable marker encodes a polypeptide conferring antibiotic resistance, said antibiotic being selected from the group consisting of actinomycin, ampicillin, chloramphenicol, erythromycin, gentamycin sulfate, hygromycin, kanamycin, neomycin, penicillin, polymixin B sulfate and streptomycin sulfate.

59. (original): The vector of claim 33, wherein said selectable marker coding sequence is a polynucleotide sequence encoding a polypeptide conferring antibiotic resistance, said antibiotic being selected form the group consisting of actinomycin, ampicillin, chloramphenicol, erythromycin, gentamycin sulfate, hygromycin, kanamycin, neomycin, penicillin, polymixin B sulfate and streptomycin sulfate.